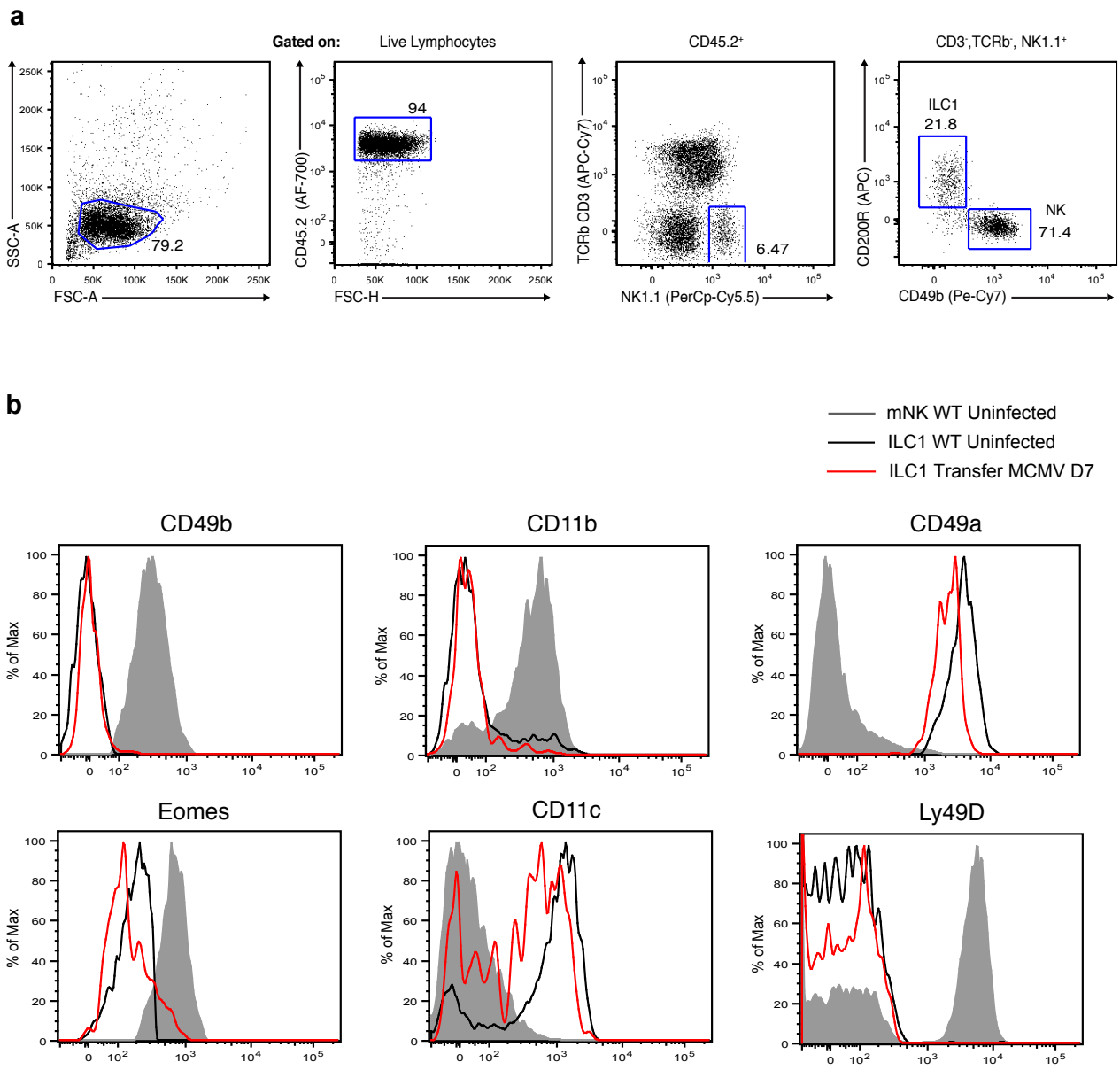
Supplmentary Figure 1

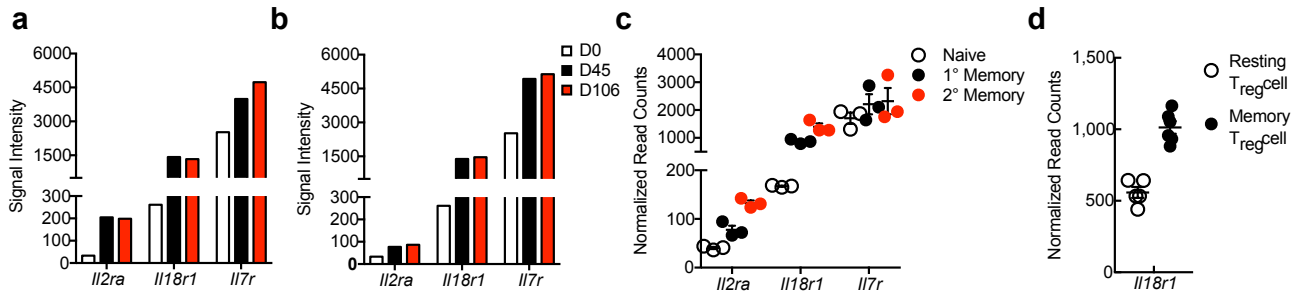


Supplementary Figure 1

Figure S1. Related to Figure 2. Phenotypic and functional stability of Liver ILC1 during MCMV challenge.

(a) Flow cytometry gating strategy for identifying Liver ILC1**. (b)** 4x10^4 liver ILC1 (Lin$^-$NK1.1$^+$ CD49b$^-$CD200r1$^+$CD11b$^-$Ly49H$^-$) were sort purified from CD45.1$^+$ mice, adoptively transferred i.v. into Ly49H-deficient CD45.2$^+$ WT hosts, and subsequently infected with MCMV. Histograms show indicated cell surface markers on liver ILC1 and mNK cells from uninfected WT mice and adoptively transferred liver ILC1 recovered 7 days PI. Data are representative of 3 independent experiments with n=3 mice per group.

Supplemental Figure 2

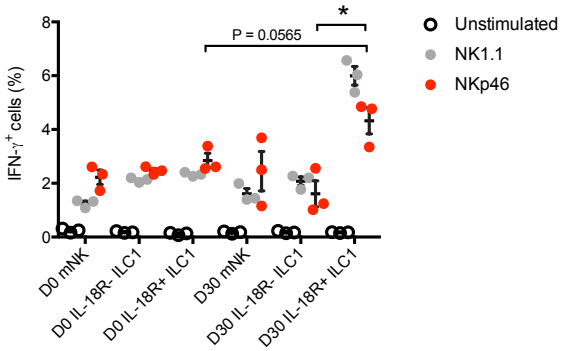


Supplementary Figure 2

Figure S2. Related to Figure 3. Memory T cell populations increase the expression of cytokine receptors following the resolution of infection.

(a,b) Graph shows average mRNA expression of indicated cytokine receptors in splenic OT-I $CD8^+$ T Cell sorted from **(a)** Listeria-OVA and **(b)** VSV-OVA infected mice at indicated time-points PI, as assessed by microarray (Data provided by Immunological Genome Consortium [54]. **(c-d)** Graph shows mRNA expression of indicated cytokine receptors by **(c)** naïve and LCMV experienced primary and secondary memory GP66-$CD4^+$ T cells post LCMV infection and by **(d)** resting and inflammation experienced memory Tregs, assed by RNA-sequencing as was previously reported [23]. Data are representative of 2 independent experiments with **(c)** n=3 and **(d)** n=5 mice per group. Data are presented as the mean ± SEM.

Supplemental Figure 3

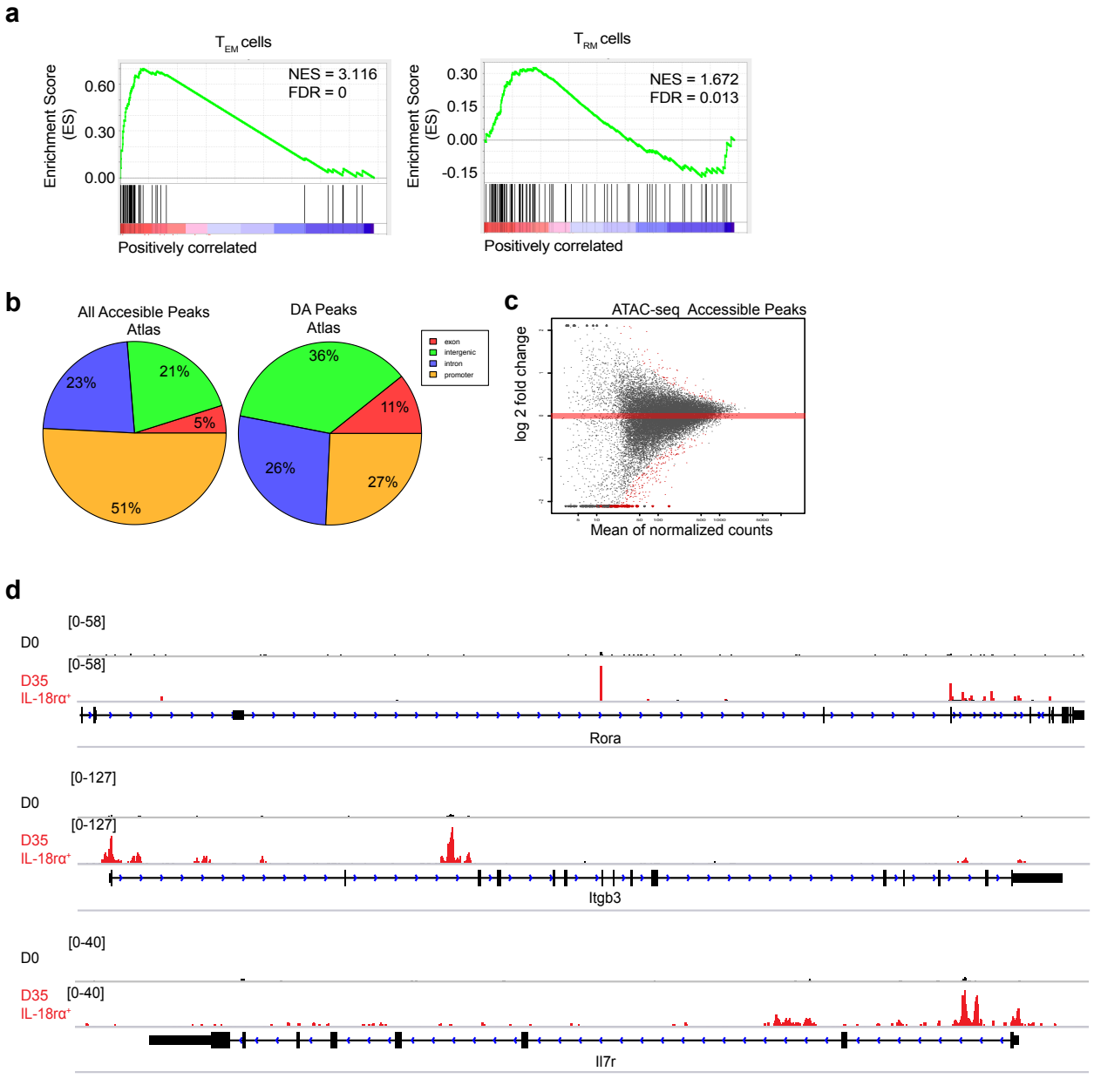


Supplementary Figure 3

Figure S3. Related to Figure 4. Il-18rα^{+} ILC1 display enhanced IFN-γ production following stimulation of activating receptors.

WT mice were infected with MCMV (i.p.) and liver was harvested and analyzed 30 days PI. Graph shows percentage of IFN-γ^{+} cells within indicated liver ILC1 populations following plate-bound stimulation with either media alone, αNKp46, or αNK1.1 plate-bound antibodies compared to uninfected mice. Data are representative of 3 independent experiments with n=3 mice per group. Samples were compared using a two-tailed Student's t test, and data are presented as the mean ± SEM (*p<0.05).

Supplmentary Figure 4

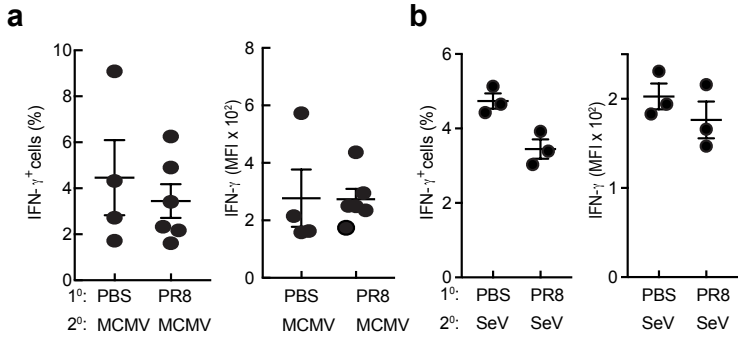


Supplementary Figure 4

Figure S4. Related to Figure S5. Memory ILC1 have distinct transcriptome and epigenomes compared to naïve ILC1.

(a) Gene set enrichment analysis (GSEA) of genes upregulated in D35 IL-18rα^+ ILC1 over D0 ILC1 from genes upregulated in effector memory T cells (T_{EM}) (left) or resident memory T cells (T_{RM}) (right) compared to naïve T cells after LCMV infection, assessed by RNA-sequencing as was previously reported[32]. (NES, normalized enrichment score; FDR, false discovery rate; NES, normalized enrichment score assessed by an empirical phenotype based permutation test assuming a null distribution). **(b)** Absolute numbers and proportion of all peaks (23016 total) and differentially accessible (DA) peaks in peak atlas (373 total, p value less than 0.2). **(c)** MA plots of differentially accessible regions (red dots) of all peak types comparing D0 vs D35 IL-18ra$^+$ ILC1. **(d)** Representative ATAC-sequencing tracks show accessible regions for *Rora, Itgb3,* and *Il7r* in naïve and memory ILC1. *Y* axis depicts normalized counts, while *x* axis displays genomic axis with scale bar. Data are representative of 2 replicate experiments with n=2 samples of n=20 mice per condition. All adjusted p values were indeed determined using DESeq2 and were two-sided.

Supplemental Figure 5

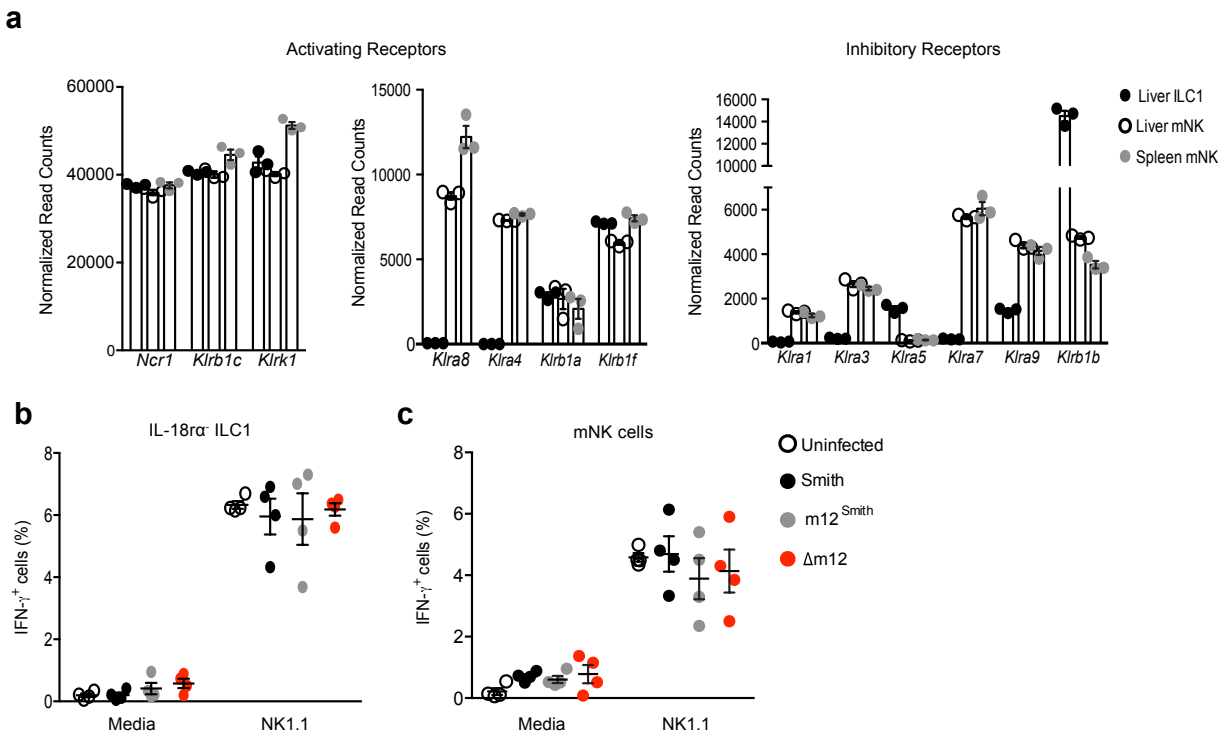


Supplementary Figure 5

Figure S5. Related to Figure 6. Induction of ILC1 memory is MCMV specific and cannot be formed by other viruses.

.WT mice were injected initially with either PBS or Influenza-PR8 intranasally (i.n.). 28 days PI, mice were subsequently challenged with either MCMV (i.n.) or Sendai virus (Sev) and analyzed 48 hours PI. **(a)** Quantification of intracellular IFN-γ staining by percentage and MFI of ILC1 at 48 hours following primary and secondary MCMV challenge in i.v. CD45 unlabeled fraction of the lung. **(b)** Quantification of intracellular IFN-γ staining by percentage and MFI of ILC1 at 48 hours following primary and secondary SeV challenge in i.v. CD45 unlabeled fraction of the lung. Data are representative of 3 independent experiments with **(a)** n=4 mice and **(b)** n=3 per group. Data are presented as the mean ± SEM.

Supplemental Figure 6



Supplementary Figure 6

Figure S6. Related to Figure 6. The MCMV-encoded protein m12 does not drive memory formation of IL-18rα- ILC1.

(a) Graph shows mRNA expression of indicated Group 1 ILC specific activating and inhibitory receptors in resting liver ILC1, liver mNK, and splenic mNK, obtained by RNA-sequencing as was previously reported [12]. **(b-c)** WT mice were infected with indicated MCMV strains i.p. and liver ILC1 were analyzed 30 days PI. Quantification of IFN-γ^+ cells within liver for **(b)** IL18rα^- ILC1 and **(c)** mNK following plate-bound stimulation with either media alone or αNK1.1 antibody. Data are representative of 3 independent experiments with **(a)** n=3 mice and **(b)** n=4 per group. Data are presented as the mean ± SEM.